Plastic-(not)fantastic? Effect of bisphenol A and its analogues on calcium homeostasis in mammalian oocytes

Bisphenol A (BPA) is an organic compound belonging to phenols, widely used in production of resins, polycarbonates, and plastics. Due to its transparency, lightness, and resistance to damage, it is used in the production of many everyday objects, e.g., plastic food containers, toys for children, CDs and DVDs, metal cans (it covers their inner surface), paper for thermal printers, cosmetics, dental fillings. Mass production of this compound contributed to its spread in the environment. BPA can be released from plastic by changes in pH and temperature, and migrate to food, air, skin, saliva, and blood. **Unfortunately, BPA belongs to xenoestrogens, i.e., synthetic compounds exerting an estrogen-like effect. Therefore, it may disrupt functioning of animal and human organisms, including negatively affecting their fertility.** Indeed, previous studies have shown that BPA reduces the quality of eggs and sperm.

The main question we want to address in this project is whether BPA, widely spread in our environment, affects the ability of mouse egg cells to generate oscillations of cytoplasmic calcium concentration in response to fertilization and, consequently, decreases female fertility. Calcium oscillations are extremely important for reproduction because they determine proper transformation of a fertilized egg into an embryo: they induce completion of meiotic division and initiation of mitotic divisions of the embryo, initiate establishment of the block to polyspermy, regulate functioning of mitochondria during fertilization and the expression of genes in the embryo. **Therefore, we also wish to investigate whether BPA-induced alterations in the pattern of calcium oscillations correlate with reduced developmental capabilities of the resulting embryos. Next, we will examine the mechanism involved in BPA action on egg cells.** We will focus primarily on the interaction of BPA with estrogen receptors present in eggs, the ERK/MAPK-dependent intracellular signaling pathway, and the influence of BPA on the amount of free reactive oxygen species and the activity of mitochondria. **Moreover, we will test whether the susceptibility of egg cells to BPA depends on the female age. Finally, we will examine how BPA analogues, bisphenol S and F (BPS and BPF), affect ability of fertilized eggs to generate calcium oscillations.** As public awareness of the potential harmfulness of BPA increases, other substances, including BPS and BPF, have been replacing this compound in plastic production. Unfortunately, BPS and BPF also display xenoestrogenic properties. Moreover, their impact on human and animal organisms is poorly understood, making their use potentially even more risky.

In our project, we will use **mouse egg cells**, as mouse is a well-established model in developmental and reproductive biology of mammals. BPA and its analogs will be administered during maturation of eggs, either *in vitro* (as an addition to the culture medium) or *in vivo* (administered to mice orally). We will work primarily with the concentrations similar to these detected in ovarian follicle fluid (*in vitro* experiments) and to estimated human daily intake (adjusted for body weight; *in vivo* experiments). In our experiments, we will use a wide spectrum of molecular and cell biology techniques.

We believe that our project will show whether – and, if so, how - BPA affects the ability of fertilized eggs to generate calcium oscillations. Our preliminary studies indicate that BPA in the concentration typical for ovarian follicular fluid (i.e., the concentration typical for the egg's surroundings under physiological conditions) administered during *in vitro* maturation of eggs shortens the duration of calcium oscillations and increases their frequency. This can potentially affect quality of the resulting embryos. Moreover, our results suggest that estrogen receptor GPR30 and ERK/MAPK signaling pathway are involved in the action of BPA. We would like to learn more about this mechanism and to investigate whether other receptors and signaling pathways are engaged in it. Our recent data also show that eggs from older females are less susceptible to BPA than eggs from young females, so we plan to find the reason for this difference. Finally, we hope to determine if BPS and BPF affect the ability of fertilized eggs to generate calcium oscillations and use this data as preliminary results for future grant applications. **Overall, we believe that our project will shed new light on the impact of BPA and its analogues on mammalian eggs, and hence female fertility**.